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CONTENTS

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THE ROYAL ENTOMOLOGICAL SOCIETY OF LONDON

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PROCEEDINGS OF THE
ROYAL ENTOMOLOGICAL SOCIETY OF LONDON

SERIES A. GENERAL ENTOMOLOGY

VOLUME 19. 1944.

GLOSSINA PALPALIS FUSCIPES BREEDING AWAY FROM WATER (DIPTERA) [1]

By T. W. CHORLEY, F.R.E.S.

So far as I am aware, there are no definite records of *Glossina palpalis* breeding more than a mile from water. Fiske (1920 : 384–389, 454) gives several instances of *G. palpalis* [*fuscipes*] occurring inland, but the maximum distance from water at which he records finding the species breeding is 1200 yards, on Mbugwe Island, where he found pupae at this distance from the water (: 384). He states (: 454) that, apart from shade and earth suitable for larviposition, "the insect appears to require nothing except that which is inseparable from an equable, tropical climate. It is believed to be pure coincidence that the insect is never found far from water, and it is confidently believed that it can exist, and that it will be found eventually to exist, in any inland localities where host animals of favoured species occur in well-sheltered areas provided with suitable breeding-places." Swynnerton (1936 : 126, 127) was evidently not aware of any other records of *palpalis* breeding inland, for he merely quotes Fiske's records and comments : "To say, therefore, that *G. palpalis* cannot inhabit wooding otherwise suitable for more than a given distance from the shore would be untrue; but under the condition of lack of suitable food animals with localised and regular habits, it cannot do so." Newstead (1924 : 118) quotes Schwetz as having found one living pupa and three empty puparia of this species in a grove 1650 yards away from water in the Kwango District of the Belgian Congo. There are several other records of *palpalis* being found in numbers away from water, but the only other record of the species breeding in such places which I have been able to find is in a paper which I have not seen : Zumpt (1937; see *Rev. appl. Ent.* (B) **25** : 125) is stated to have found *G. palpalis palpalis* breeding "at a distance from water" in conditions of high humidity. The fact that *G. palpalis fuscipes* has now been found breeding several miles from water confirms Fiske's belief, and is of considerable interest.

The breeding-places now recorded were discovered in Busoga District, Uganda, and were in dense, humid, forest, broken into by numerous elephant-tracks. One of these breeding-places (at Nandegeyi) was at a distance of four miles from the nearest permanent water and about five miles from Lake Victoria,

[1] Published by permission of the Director of Medical Services, Uganda.

and breeding was taking place here during the dry season. At this particular place the only water for many miles around, during the dry season, is in a small water-hole which was used by the local people as a water supply before the evacuation of the area owing to sleeping sickness. The period during which all but one of the pupae were collected (24th March to 14th April 1943) should have been the time of the long rains, but 1943 was a year of drought and there was no considerable amount of rain until near the end of April. The normal dry season is from December to February inclusive, and in 1943 this period was even drier than usual.

My attention was first drawn to the possibility of *G. palpalis* breeding far away from water by the unusual density of this species in areas several miles from the lake-shore, and the high proportion of males in these areas. A search for pupae was begun in February 1943, and the first pupa was found on the 26th of the month. It lay under a fallen tree-trunk in undulating forest country on a ridge called Busakira; ten days later a male *G. palpalis fuscipes* emerged from this pupa. Arrangements were then made to undertake a further search at various points in a large portion of the South Busoga Sleeping Sickness area. This later search extended over an area of about 25 by 10 miles, and breeding was found to be widespread throughout the area searched. Except at Nandegeyi, less attention was paid to the factor of the distance from the breeding-places to small water-holes, partly because the main object was to discover the maximum distance inland to which breeding extended, and partly because the breeding-places found were in no case around such water-holes. In the whole area examined there is, during the dry season, no water nearer than the lake except in such water-holes, which are far from numerous. During the rains there are swampy areas in shallow valleys, most of which are not traversed by streams, but these swamps rarely persist for more than two months, and are not breeding-places of *palpalis* because of the complete absence of suitable shade and suitable soil-conditions. Some of them apparently serve as pathways for *palpalis* during the rains, and tsetse travelling along them may reïnforce the permanent population of the inland haunts. Of the seventeen areas in which breeding was found, some of which included several breeding sites, the one most distant from Lake Victoria was twelve miles from the lake.

Breeding was often on a large scale, one such site producing 14 living pupae and 39 hatched puparia in less than a quarter of an hour. From seven breeding sites in one area of about two miles square, 17 pupae and 96 hatched cases were obtained. The breeding was certainly of long standing, because many of the empty cases found were very old. That many of the pupae had been deposited during the dry season is proved by the period prior to hatching: the vast majority of the flies hatched from the puparia in three or four weeks after collection, a number took five weeks, and two took 48 days each. Since Symes and Southby (1938 : 23) found that the pupal period of *G. palpalis* [*fuscipes*] varies in natural conditions (in an area in Kenya which is only about 25 miles away from that in which the pupae were found in Busoga) from 43 to 53 days, and since the maximum pupal period recorded in Uganda is 55 days, it is evident that most of the puparia had been deposited in the period from January to March, in the heart of the dry spell.

Type of Country in which Breeding Areas were Found.

A large part of southern Busoga is covered with dense rain-forest, but *Glossina palpalis* was found breeding only in those areas of forest in which a

network of elephant paths provided a suitable environment (mixed light and shade) for the adult fly. Where elephants are accustomed to feed and rest in the forest, conditions are exceptionally favourable for the fly, owing to the destruction of undergrowth and the pulling down of trees by these animals, the forest being thus opened up and sunlight allowed to enter. No adult *palpalis* were found, nor was there any evidence of the species breeding, in places which had not been visited by elephant at some time during the previous year.

The forests consist mainly of deciduous trees, some of those commonly met with being *Chlorophora excelsa*, *Pseudospondias microcarpa*, *Albizzia coriaria* and several other species of this genus, *Ficus* spp. and *Cordia* spp.; *Phoenix reclinata* also occurs in some parts. The undergrowth in the forests is never really dense, and *Amomum granumparadisii* forms a very large part of it, together with *Dracaena ugandensis*. In some places *Sansevieria* grows very profusely and where this plant occurs very little other undergrowth is present.

Types of Breeding Sites Chosen by the Fly.

All the breeding sites found were under fallen trees or at the bases of standing trees. For some years there has been much felling for timber of *Chlorophora excelsa* in some of the forests examined, and the portions not removed to the saw-mill form very favourable breeding sites for *palpalis*. The usual practice is to leave the crown of the tree, comprising large branches and a small part of the trunk, in the forest; and as termites are reluctant to attack the very hard wood of this tree, the remains persist for long periods. In areas where no tree-felling has taken place, suitable breeding sites are provided by trees pulled down by elephants or blown down by the wind. Provided that conditions of light and shade were suitable and the ground was not too wet, pupae were found under about two-thirds of all the fallen trunks examined, in one instance 14 pupae and 39 empty puparia under one such log. On three occasions pupae of *G. palpalis* were accompanied by those of *G. pallidipes*, and in one instance the latter species occurred by itself. In addition, pupae of *pallidipes* were found under one fallen trunk in the open, where shade conditions were not suitable for *palpalis*. Adults of *pallidipes* were even more abundant than those of *palpalis*, but breeding conditions were not optimum for the former species.

Breeding sites at the bases of standing trees were much rarer. Many apparently suitable sites of this type were searched, but pupae or puparia of *palpalis* were only found in five of them. Of these, two were among the buttresses of large trees (*Pseudospondias microcarpa*) and the other three at the bases of large specimens of *Chlorophora excelsa*, either close to the tree or (in one instance) at a point where a clump of *Dracaena ugandensis* formed a canopy a few yards from the tree but within the area shaded by its crown. This clump was the lair of a spotted hyena.

Food Supply Available to the Fly.

Besides elephant, bushbuck (*Tragelaphus scriptus* subsp.), bush pigs (*Potamochoerus porcus* subsp.) and buffalo (*Syncerus caffer* subsp.) are all very common in the forests examined. Waterbuck (*Kobus defassa* subsp.) and brown duiker (*Sylvicapra grimmia* subsp.) are plentiful in the area, but seldom penetrate deeply into the forest, preferring to lie up near the edge of the forest or in

thickets outside it. Leopard and spotted hyena (*Crocuta crocuta*) are occasionally met with in the forest, and hippopotami occasionally wander into it from Lake Victoria. Large fruit-bats and porcupines (*Hystrix galeata* subsp. and perhaps also *Atherura turneri*) are present in the forest but are rarely seen. Monkeys are usually abundant in the forests near Lake Victoria, but only two individuals were seen during the search for breeding-places of *palpalis*. Man is now excluded from the area by law, but before the evacuation the local Africans frequently made excursions into the forests in search of game, fruit, and hut-building material, and there is certainly still a certain amount of trespassing into them, traces of recent human activities being found during the survey.

Turning to reptiles, *Varanus* (a favourite food of *palpalis* on the lake shore) is present. Forests of the type examined usually contain numerous snakes, but none was actually observed.

There is no doubt that the fauna of these forests provides an ample food supply for *G. palpalis*, as is the case in all the forests in Uganda which we have examined.

Summary.

Glossina palpalis fuscipes has been found breeding, in the Busoga District of Uganda, in places far removed from any permanent water. In one instance the nearest water of any kind was four miles away; in others breeding was taking place at distances up to twelve miles from any permanent water except small water-holes. The area in which the breeding-places were found, and the sites in which breeding occurred are described, and the food supply of the tsetse is discussed. The exceptional distance from water at which breeding of *palpalis* occurs in Busoga is thought to be due to the very humid conditions in the forest. Swynnerton's suggestion that food supply is the factor which normally limits *palpalis* to the near neighbourhood of permanent water is probably incorrect. In Uganda suitable conditions of shade and humidity seldom occur far from permanent water, and where these conditions are not found *palpalis* is absent, no matter how ample the food supply.

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A MASS CATCH OF CABBAGE WHITES BY SUNDEWS

By Prof. F. W. Oliver, F.R.S.

In the course of visiting a number of islands in Sutton Broad, Norfolk, on 4th August, 1911, our boat drew alongside an island towards the end of that arm of the broad which stretches in the direction of Sutton village. The surface of the island, which was about two acres, was a perfectly level sward, some thirty inches, perhaps three feet, above the then water-level. Everywhere upon this sward were fluttering white objects, which merely fluttered and made no progress. They were cabbage white butterflies, *Pieris rapae*. The reason for their getting nowhere was that they were anchored to the sticky leaves of the sundews which constituted the sward.

These sundews were *Drosera anglica*, a small tufted plant, a few inches high. The blade at the end of each leaf is densely set with little glandular hairs ending in tiny, spherical bulbs, from which exudes a sticky, glistening excretion that attracts insects. Once caught, the insects remain stuck, whilst the hairs ultimately converge on their bodies and pour out a digestive fluid which dissolves their proteins—these in turn being absorbed into the plant.

Each *Drosera* plant had from four to seven Cabbage Whites adhering in this way, mostly still alive and struggling to be free. Making several counts, it appeared that seventy Whites to the square foot of ground was the average. As the number of square feet in an acre is 43,560, this means that the *Drosera* plants of our two-acre field had recently captured some six million Cabbage Whites, and were in process of dissolving and digesting them.

The sun rose that day at about 4.30 a.m. and it was now 1.30 p.m. (G.M.T.). From the condition of the insects it was evident that they had arrived on the field and been captured that morning. Suppose the swarm to have arrived soon after daylight—this would give an outside limit of nine hours for the *Drosera* to have been at this banquet. In a number of cases (perhaps 30%) parts of the bodies were already in obvious process of solution.

It is well ascertained that Cabbage Whites travel about in vast swarms and it may be reasonably supposed that this swarm had come in early from the sea (it is, I understand, accepted by entomologists that these insects often migrate in this way from the continent of Europe). The coast (at Palling) is just four miles from this part of Sutton Broad. Passing inland and travelling over our little island, they had been attracted by the *Drosera*. One may imagine that the head of the flight settled on the plants till no more "seats" remained, and that the rest of the swarm pursued its course in the hope, perhaps, of finding unoccupied *Drosera* further on. Anyway, there were no free Whites still hovering disconsolately; nor during the time I was on the island did I notice even a single specimen regaining its freedom.

As to the *Drosera*, I remember they were unusually large plants, and more brightly coloured with crimson sap than is usual. It seemed probable, although the banquet was still in its earlier stages, that the *Drosera* were already showing a favourable reaction. Being bound to the party I was unable to remain and watch the sequel. The year 1911 was outstanding for its prolonged drought.

Specimens were collected at the time, and I remember for thirty years there was one in a glass case showing the insects *in situ*—a museum specimen shown to generations of students at University College, London. Whether it has survived successive "blitzes" and evacuation I cannot say.

DUCTEOLES OF THE SECRETORY CELLS IN THE SPERMATHECA OF *PSILA ROSAE* FABRICIUS (DIPTERA, PSILIDAE)

By G. Pontecorvo, Ph.D.

(*Department of Zoology, Glasgow.*)

While investigating the occurrence of polytene chromosomes in the tissues of Diptera, I made the following casual observation on the histology of the spermatheca of *Psila rosae.*

The spermatheca is cylindrical with a central tube, in which spermatozoa are stored, thickly cuticularised and surrounded by large secretory cells. These have a nucleus, with polytene chromosomes, placed distally in the cell relative to the lumen of the spermatheca. A large vacuole, in which secretion collects, occupies the greater part of the proximal region of the cell.

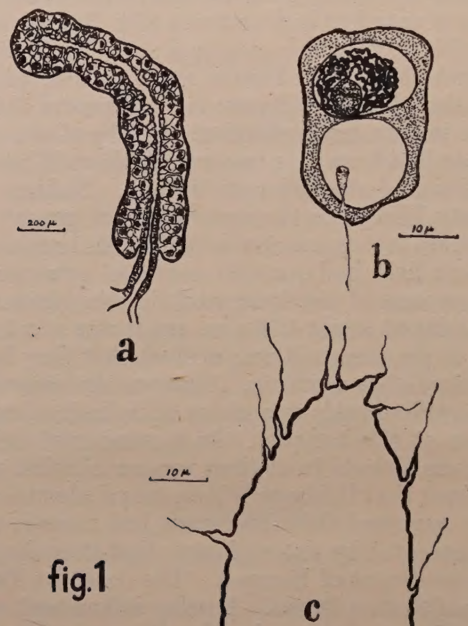


Fig. 1.—*a*, Diagrammatic drawing of the spermatheca of *Psila rosae*; *b*, Secretory cell showing the funnel-like opening of the ducteole into the vacuole; *c*, Tip of the spermathecal lumen showing openings of ducteoles into its cuticularised wall.

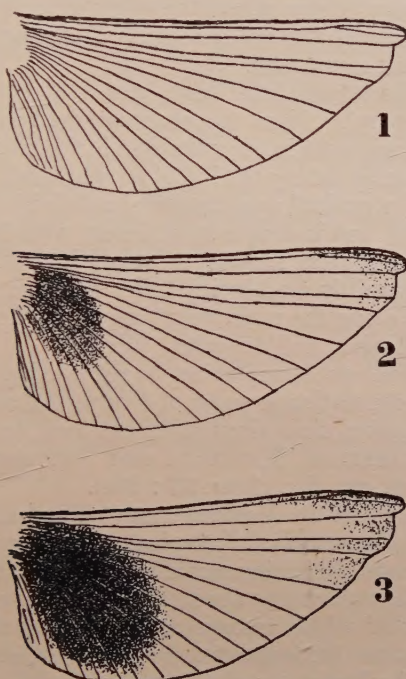
The interesting feature is that a very thin ducteole, almost certainly chitinised, connects the vacuole with the central lumen of the spermatheca. Each cell has its own independent ducteole : I have followed many throughout their whole course and never found any anastomosis. The length of the ducteole depends on the distance of the respective cell from the central lumen, and may be even greater than 200 $m\mu$; its diameter is uniform, about 0·3 $m\mu$. At both ends, into the vacuole of the cell and into the wall of the lumen, the ducteole opens out into a sort of funnel with mouth up to 3 $m\mu$ in diameter.

The casual observation of these elusive ducteoles was, no doubt, facilitated by my working with whole mounts flattened or crushed and overstained with acetic orcein.

CHANGES IN WING PIGMENTATION DURING THE ADULT LIFE OF ACRIDIDAE (ORTHOPTERA)

By E. D. Burtt, Ph.D., F.R.E.S., and B. P. Uvarov, C.M.G., D.Sc., F.R.E.S.

While collecting Acrididae in Tanganyika Territory, one of us (E. Burtt) observed that *Mesopsis laticornis* (Krauss 1877) occurred in two colour forms, one with perfectly hyaline hind-wings and another with the wings bearing a large black spot at their base; the latter has been described as var. *infuscata* Krauss. The two extreme colour forms were, however, found to be connected by intermediate ones, and observations throughout a season showed that the



Figs. 1–3.—1, Hind-wing of *Mesopsis laticornis* (Krauss), male, in June. 2, Ditto, in November. 3, Ditto, in December. Somewhat enlarged; venation shown diagrammatically, without cross-veins.

hyaline forms are prevalent in the early dry season but are replaced by the infuscate ones at the beginning of the next rainy season. These observations led Burtt to suggest that the difference between the two forms is due to a gradual deposition of pigment in the wings of an originally clear-winged individual (Uvarov 1943, *Eos* **19** : 78). This explanation required direct experimental corroboration, which has now been obtained.

On 12th June 1942, Burtt collected a series of adults of *Mesopsis laticornis* of both sexes from a population of a small marsh just below the Sleeping Sickness Research Laboratory at Tinde, 20 m. S.W. of New Shinyanga, Tanganyika Territory. All specimens had perfectly hyaline wings (fig. 1) showing no trace

of dark colour even on the veins, which were whitish. The insects were kept in a cage in the laboratory and fed on millet leaves. A sample examined on 14th November showed that most individuals developed a faint, but distinct, yellowish tinge of the membrane of the hind-wings, while the main veins became brownish in their basal sections, and the membrane here became distinctly infumated, forming a small spot, while lighter infumation of the membrane was perceptible in the apical part of the wing (fig. 2).

Another sample taken from the same cage on 18th–29th December showed the wing pattern of the f. *infuscata* fully developed. Most of the wing veins and veinlets were brown to black, and the membrane was lightly infumated throughout the wing, more distinctly so in the apical portion and along the costal edge, while the basal spot became large and was of shiny brownish-black colour (fig. 3). This colour of the spot is due to the pigment deposited in the membrane, and it is more intense round the edges of each cell, while the middle is paler; the cross veins and some of the main veins remain pale and stand out sharply against the dark membrane.

The series of samples taken at the dates stated above is now in the British Museum (Natural History). Thus the observations leave no doubt that the deposition of the black pigment in the veins and in the membrane of the wings is a gradual process occurring during the life of an adult individual. This process may be analogous to the gradual appearance of a definitive adult pattern in the recently moulted adults, but it occurs in an insect with fully hardened integument and extends over several months of adult life. The pigment must be a metabolic product (possibly a melanin) and it must be transported from the body by blood flowing along the main veins, spreading from them to the membrane, probably by diffusion, and deposited there.

Thus, the observations on *Mesopsis* can be regarded as an indirect, but conclusive, proof of blood circulation in wings of Acrididae, a fact not yet recorded, although one to be expected since circulation has been definitely shown to take place in wings of Blattidae, Mantidae and Tettigoniidae (Yeager and Hendrickson 1934, *Ann. ent. Soc. Amer.* **27** : 261).

The pigment metabolism in *Mesopsis* may be connected with the sexual maturation, since many Acrididae of tropical African grasslands remain adult throughout the dry season, becoming sexually mature at the onset of the rains. A very typical example is offered by the Red Locust, *Nomadacris septemfasciata* (Serville 1839), adults of which emerge at the end of the rainy season, but remain sexually immature throughout the dry season, and mature only after several months with the beginning of the next rains (Uvarov 1933, *Bull. ent. Res.* **24** : 419), when the hitherto colourless hind-wings become bright rose.

The implications of the changes in the pigmentation described above are as follows :—

1. The pigmentation of an adult Acridid may be subject to considerable changes, suggesting the continuation of metabolic processes and of circulation even in fully hardened integuments and in wings.

2. Great caution should be exercised in regarding colour and pattern differences in Acrididae as taxonomic characters, since only some of them may be attributed to genetical causes (Rubtzov 1935, *Bull. ent. Res.* **26** : 499) while others represent responses to the environment of the nymphal stage (Faure 1932, *Bull. ent. Res.* **20** : 671) and still others are due to metabolic processes in the adult.

LARVAE OF THE BRITISH TRICHOPTERA. 20

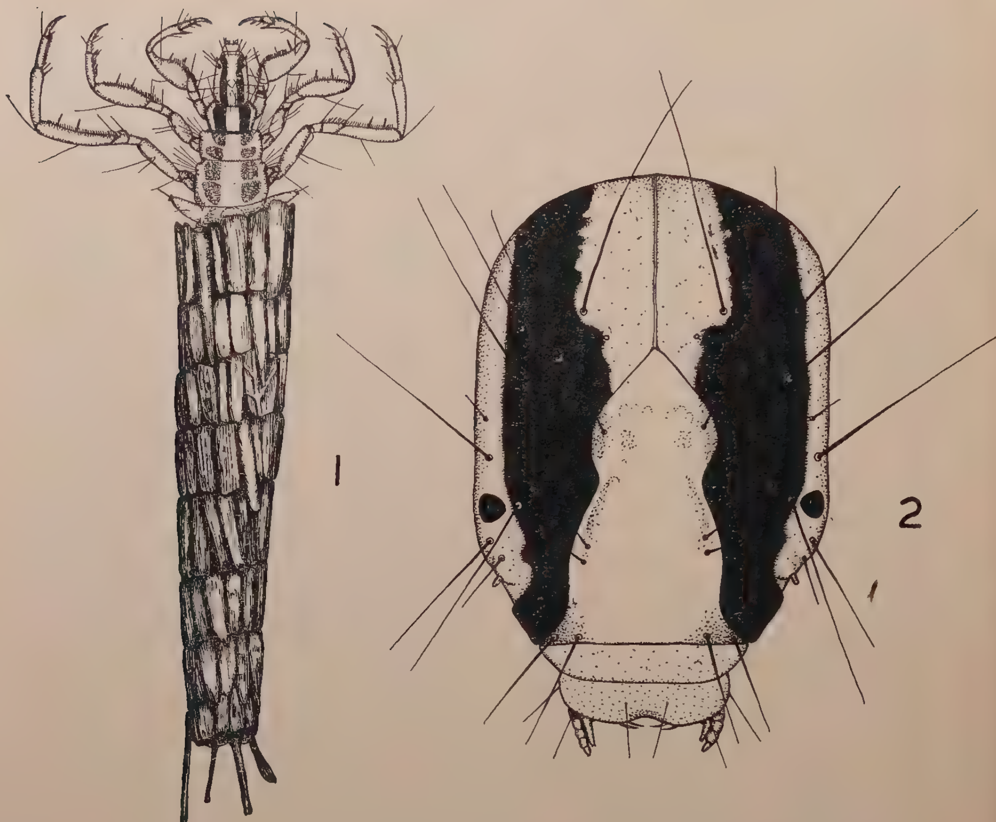
By N. E. Hickin, Ph.D., F.R.E.S.

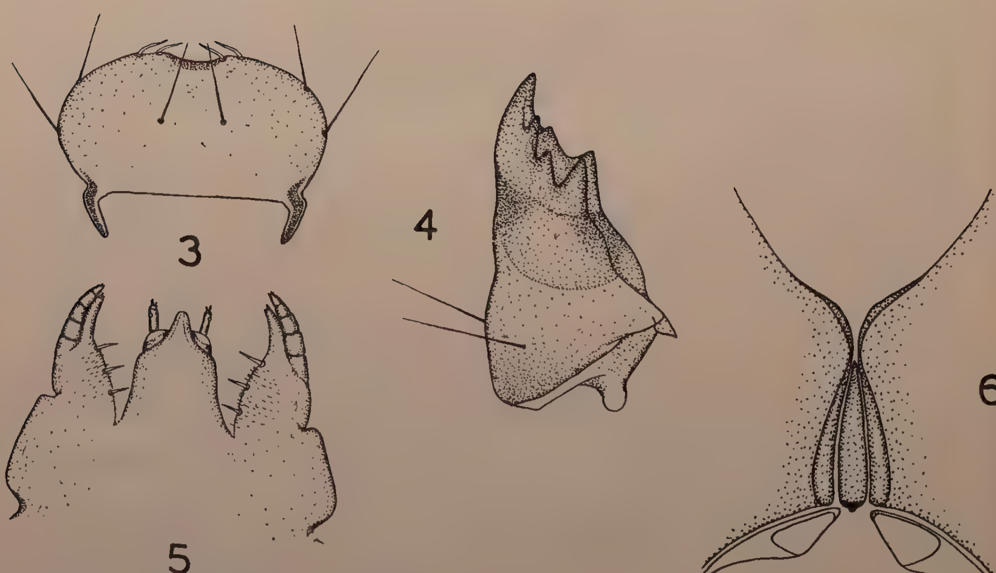
Neuronia ruficrus Scopoli (Phryganeidae).

My first experience of this species was in 1937 when I collected three larvae from a pool in Broadmoor Wood, Rubery, Worcestershire (about 650 feet above sea-level). These larvae were successfully reared to the adult stage. Each year since I have taken some adults, always resting on stems of marginal water plants, but although I collected extensively each spring in this locality I failed to obtain more larvae. In September 1943, however, Mr. W. E. China very kindly showed me the pool in Brathay Quarry (about 200 feet above sea-level) near Wray Castle, Lake Windermere, where he and Mr. D. E. Kimmins had taken a series of the adults at the beginning of June 1942. The larvae were quite plentiful and I collected a number. They were walking over the decaying leaves which formed the floor of the pool. Then on 10th October I revisited the Broadmoor Wood Pool and was successful in collecting a dozen larvae. The pool at Broadmoor Wood, Rubery, and this pool at Brathay Quarry have several common characters. Both are surrounded by a dense growth of trees, contain much decaying foliage, and much thick black mud at one side, and in parts are fairly deep. Lestage in Rousseau states that the larvae are crepuscular or nocturnal. My experience leads me to believe that the larvae spend the winter in deeper water, or covered with leaves, as they are plentiful in autumn, where they occur, but in spring have disappeared, while at the beginning of June the adults are plentiful again.

This is perhaps a convenient place to note that the neck and prothorax of the adult when freshly emerged are bright yellow in colour, making the insect very striking in appearance.

Larva (fig. 1). Betten's term "suberuciform" for larvae of Phryganeidae is an appropriate one; there is a fairly deep intersegmentation of the abdominal segments. Larvae of *N. ruficrus* watched in an aquarium show many characters associated with campodeiform larvae. They are very agile and often pounce on larvae of *Phryganea striata* twice or three times their size, locking the prothoracic legs around the body of the victim, which they steadily devour. The head is somewhat depressed but not approaching the truly orthocentrous condition. The head and pronotum are closely applied together, and it is doubtful if the head enjoys any movement independent of the pronotum—laterally or vertically. A striking feature of the larva is a pair of dark, longitudinal, parallel bands extending from the anterior margin of the genae over the sclerotised pronotum, continued over the unsclerotised meso- and metanota and over the abdomen almost to the anal segment. In Ulmer's description the longitudinal bands are stated to extend to the first abdominal segment—often much farther posteriorly. So that although my larvae from both localities were heavily marked, it is possible that some may be lighter than others. Length of larva 20–22 mm., width 4 mm. *Case* (fig. 1) of small pieces of cut roots and pine needles arranged spirally in about 6–7 whorls, open at both ends, anterior end larger, length 35 mm., width 5·5–7 mm. The half-grown larvae can reverse their position in the case quite easily. *Head* (fig. 2). Light golden yellow with heavy marking. Very fine sculpturing is apparent if the head is cleared with clove oil after dehydration with alcohol. Genae parallel. Two wide, parallel, black bands run longitudinally over the genae (these are the bands which are continued throughout the length of the body). Marking on the clypeus is





FIGS. 1–2.—*Neuronia ruficrus.* 1, Larva and case. 2, head.
FIGS. 3–6.—*Neuronia ruficrus.* 3, Labrum. 4, mandible (left). 5, maxillae and labium. 6, gular sclerite.

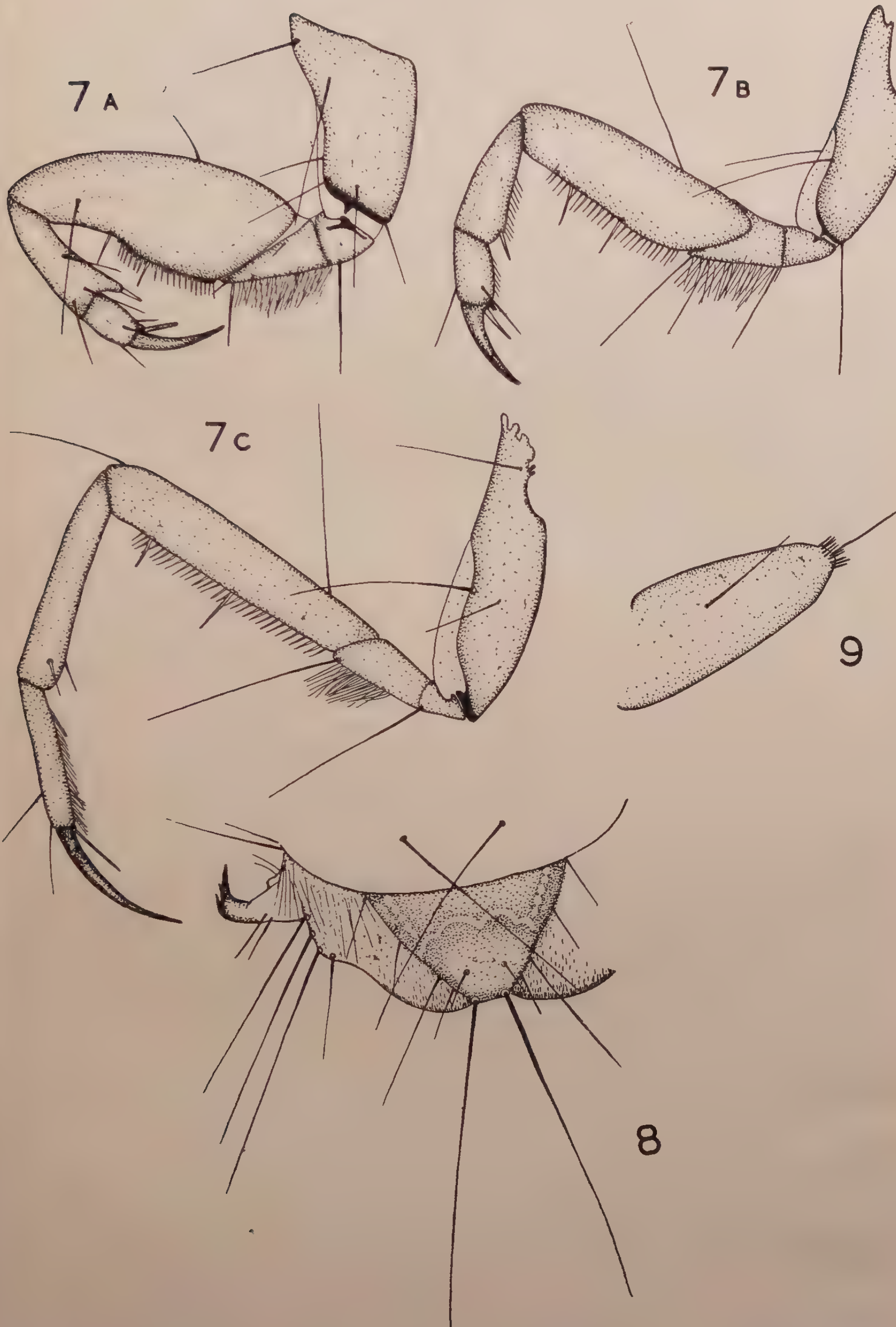


FIG. 7.—*Neuronia ruficrus.* a. Prothoracic leg. b. Mesothoracic leg. c. Metathoracic leg.
FIG. 8.—*Neuronia ruficrus.* Ninth segment of abdomen.
FIG. 9.—*Neuronia ruficrus.* Lateral protuberance on first abdominal segment.

variable but there is no median dark mark. In fig. 2 the marking is characteristic of Brathay Quarry specimens. The Broadmoor Wood specimens had lunate black marks near the lateral margins of the clypeus. Antennae are small. Eyes situated anteriorly. *Mouth parts.* Labrum (fig. 3) light yellowish-brown, elliptical, excised at centre of anterior margin and with straight transverse posterior margin. A peg-like sclerotised projection at each posterior angle. A pair of inwardly-curved yellow spines on each side of the anterior excision. Three pairs of black bristles on the dorsal surface, and three pairs of short blade-like yellow spines on the ventral surface with some short hairs. Mandibles strong (fig. 4) with three subsidiary teeth on the upper cutting edge and two on the lower. Teeth of the right mandible not so prominent as those of the left. No brush of hairs on the inner face but two bristles situated on the outer face. Maxillary palp (fig. 5) five-segmented and sclerotised but proximal segment incompletely sclerotised on the inner face. Maxillary lobe almost as long as the palp. Three inwardly-directed yellow spines on base of maxilla with some short hairs. Labial palps of two segments, the proximal one bulbous, the distal long with some sensory papillae at the tip. A sensory pit on the distal segment slightly removed from the tip is quite prominent. Labium conical, spinneret pronounced. Gular sclerite (fig. 6) long and narrow, heavily sclerotised at the anterior end. Adjacent areas of the genae folded and thickened. *Thorax.* Pronotum yellow, sclerotised with longitudinal black bands. Meso- and metanota unsclerotised, greenish-grey with continuation of black bands. In each segment each band is divided transversely into two areas, each of which is traversed by a row of light spots. Prosternal horn present, not large, apparently bent anteriorly. A small protrusion is situated on the anterior lateral vertices of meso- and metathorax, each furnished with a bunch of bristles. Pro- and mesothoracic legs short, the former deeper and slightly longer than the latter. Metathoracic legs long. In each case only the distal segment of the trochanter is hairy and ventral edge of femur has two large spines (fig. 7 a, b and c). *Abdomen.* Deep intersegmentation, greenish-grey with dark grey parallel bands continued from head and thorax. Widest at about the second segment. Lateral and dorsal protuberances on the first abdominal segment are clearly defined and white in colour. The dorsal protuberance is sharply pointed and is devoid of bristles, lateral protuberances have a bunch of sensory bristles and a long spine at the tip with a further bristle situated laterally (fig. 9). First abdominal segment narrow. Dorsal margin of ninth segment (fig. 8) shield-shaped with characteristic pattern and with two very long bristles on posterior margin. Long filiform gills present, posterior lateral gills in each segment covered with fine black hairs. Two ventral gills are present on the first abdominal segment, six gills on the second to the seventh segment (the posterior ventral gill may be absent from the seventh), and two gills on the eighth segment (of these the dorso-lateral gill may be absent). Anal claws with a pair of auxiliary claws.

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THE STRUCTURE OF THE ANTENNA OF *APHIS* (*DORALIS*) *FABAE* SCOPOLI, AND OF *MELANOXANTHERIUM SALICIS* L. (HEMIPTERA), AND SOME EXPERIMENTS ON OLFACTORY RESPONSES

By Margaret G. Jones.

(*Department of Zoology, Cambridge.*)

Very little work has been done either on the behaviour, or on the structure, of the sense organs of the Aphididae. Flögel (1905) described the structure of the sense organs of *Aphis ribis* L., and *A. platanoides* Sch., and the proximal course of the antennal nerve in *Pemphigus bursarius* L. was traced by Pflugfelder (1937). Davies (1935) carried out some experiments on the influence of humidity and wind currents on *Myzus persicae* Sulz.

The present work is concerned with the structure of the antennae of the alatae viviparae of *Aphis* (*Doralis*) *fabae* Scopoli, and with experiments on the olfactory responses of the alatae viviparae. The antennae of a larger, more primitive aphis, *Melanoxantherium salicis* L., were also studied for comparison, and sections of the antennae of *Macrosiphum pisi* Kaltenbach and *Myzus persicae* Sulz. were cut.

1. The structure of the antenna of the alatae viviparae of *A. fabae* and *M. salicis*.

Technique.

For the external examination of the cuticular parts of the antennae, alatae of *A. fabae* were treated with 70% alcohol and 70% lactic acid on a water bath, cleared in 75–95% phenol, and then mounted in Berlese's fluid.

The internal structure was examined in transverse and longitudinal sections of 5μ which had been embedded in paraffin wax with a melting point of 58° C. This relatively hard wax reduced the tearing of the chitin which occurred when a lower melting point wax was used. The majority of the antennae were removed from the heads of fresh aphids and the tips of the sixth joints were cut off to enable the fixative to penetrate the internal tissues. In some cases, both the head and antenna were fixed. Several fixatives were used :—alcoholic Bouin, Petrunkevitsch's mixture 2 (Petrunkevitsch 1933) and Zenker's fluid (McClung 1937). All gave good results, but in most cases the antennae were fixed with alcoholic Bouin. The specimens were dehydrated to 95% alcohol and then put into three changes of methyl benzoate containing 1% celloidin for 24 hours. Then two changes of benzole were used as the clearing agent, and after two hours the antennae were transferred to a mixture of paraffin and benzole for 3 hours, and then to three changes of paraffin wax. After cutting, the sections were stained with Heidenhain's iron haematoxylin alone (Eltringham 1930) or together with Orange G, or with Mallory's Triple Stain. Phosphotungstic acid haematoxylin was employed when Zenker's fluid had been used as the fixative.

External Structure.

The antennae of the alatae viviparae of *A. fabae* are six jointed, but, as in all insects, only the first two joints have associated muscles, so that the whole

flagellum (3rd, 4th, 5th and 6th joints) moves in the same direction. All the joints bear a number of hairs (fig. 1), and at the tip of the 6th joint is a group of 2–3 small hairs. The 3rd, and sometimes the 4th, joint always bears a varying number of placoid sense plates or sensoria (fig. 1). The number of sensoria on

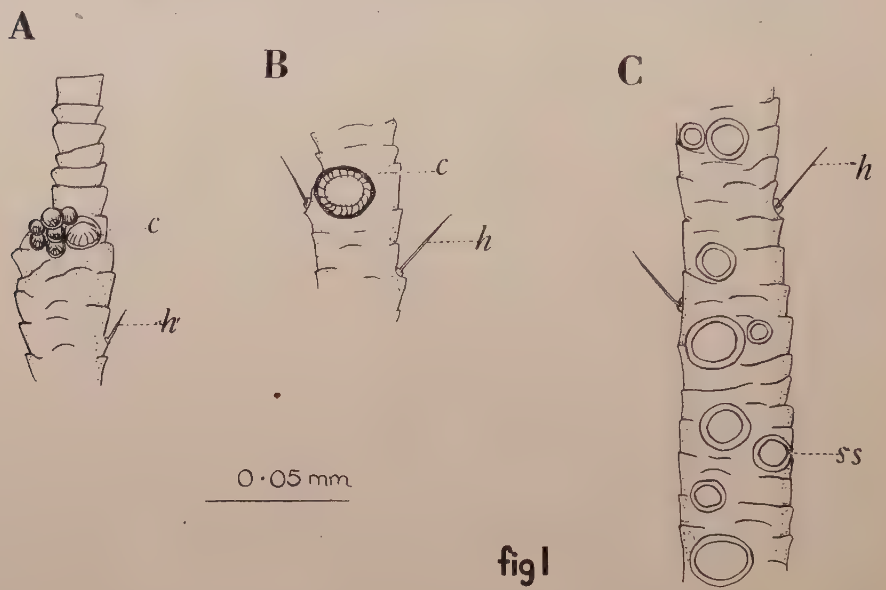

FIG. 1.—Structure of antenna of *A. fabae*. A, base of 6th joint to show primary sensorium (rhinarium); B, 5th joint to show primary sensorium; C, 3rd joint to show secondary sensoria and hairs. c = cuticular hair, h = hair, ss = secondary sensorium.

the antennae of the sexuparae is greater than that of the spring or summer forms. In the male, there are sensoria on the 3rd, 4th, and 5th joints. Table 1 gives an average figure for the number of sensoria on the different alate forms.

TABLE 1.

The number of sensoria on the 3rd, 4th, and 5th antennal joints of *A. fabae*.

Joint	Spring alatae	Summer alatae	Autumn alatae	Males
3rd	17	15	21	38
S.E.±	0·31	0·29	0·29	0·52
Number counted	121	150	152	97
4th	0·384	1	5	23
S.E.±	0·086	0·17	0·203	0·46
Number counted	91	128	155	91
5th	0	0	0·27	12
S.E.±	—	—	0·049	0·35
Number counted	>100	>100	131	86

On the distal end of the 5th joint is a placoid sense plate (primary sensorium, rhinarium) (fig. 1), which differs from the secondary sensoria by the presence of

a circle of cuticular hairs around the sense plate. On the 6th joint, at the base of the thin terminal region, is a group of 7 small sense plates, each with a ring of cuticular hairs (fig. 1). These primary sensoria are present in all the instars

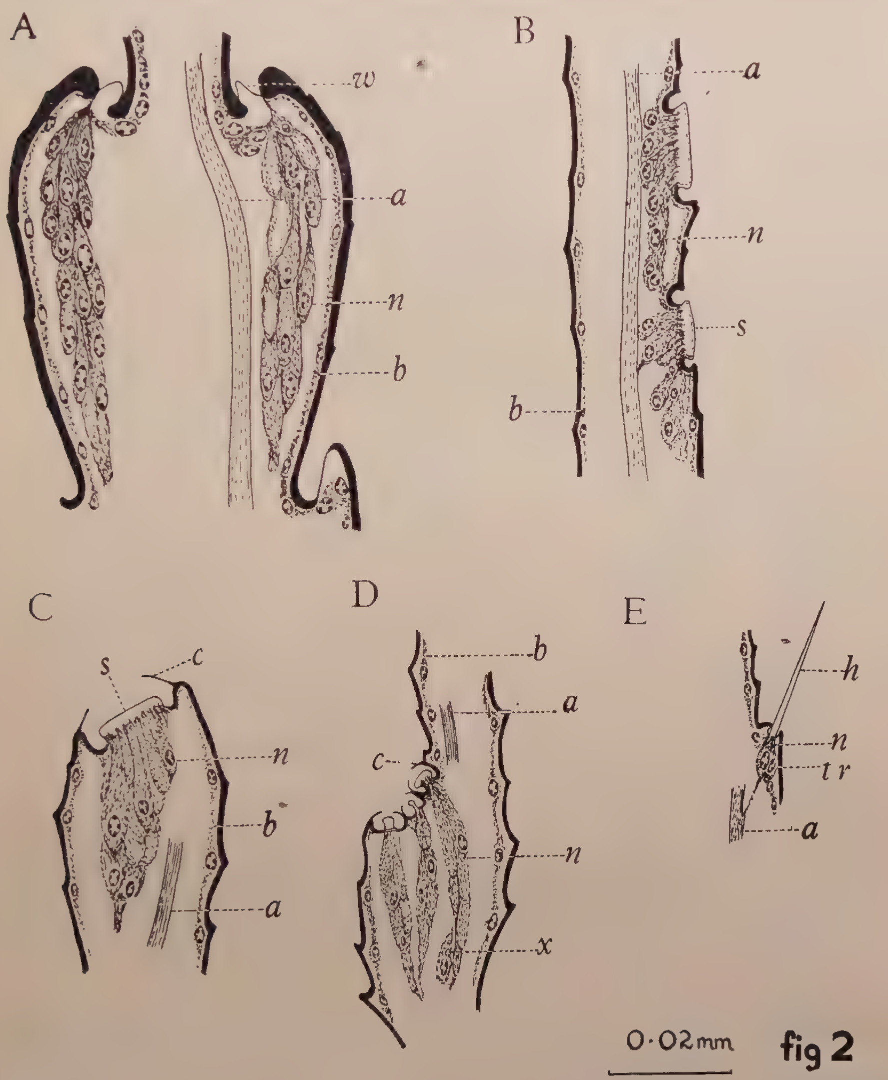

FIG. 2.—Structure of antenna of *A. fabae* as seen in longitudinal section. A, 2nd joint to show Johnston's organ; B, 3rd joint; C, 5th joint; D, base of 6th joint; E, a hair. a = antennary nerve, b = hypodermis, c = cuticular hair, h = hair, n = sense cell, s = sense plate, tr = trichogen cell, w = interarticular membrane, x = sense cell from another sensorium.

of the alatae and are found on all forms of the aphis, while the secondary sensoria are confined to the alatae.

The antennae of the apterous forms are similar, but without secondary sensoria.

The antenna of *M. salicis* is essentially the same in structure, but the

relative proportions of the joints are different, also the secondary sensoria are relatively smaller in size than are those of *A. fabae.*

Internal Structure.

The internal structure of the antenna of *A. fabae* is simple, and can readily be understood from figs. 2, 3 and 4. A single layer of living cells, the hypodermis, with evident nuclei, lies beneath the cuticle. The nuclei of these hypodermal cells often lie beneath the ridges of cuticle. Running through the antenna to the tip of the 6th joint is the antennary nerve, which is more or less centrally placed in the 1st and 2nd joints, but lies slightly to one side in the more distal joints. Pflugfelder (1937) has shown that there is only one nerve leaving the brain in *Pemphigus bursarius* L., which gives off branches to the

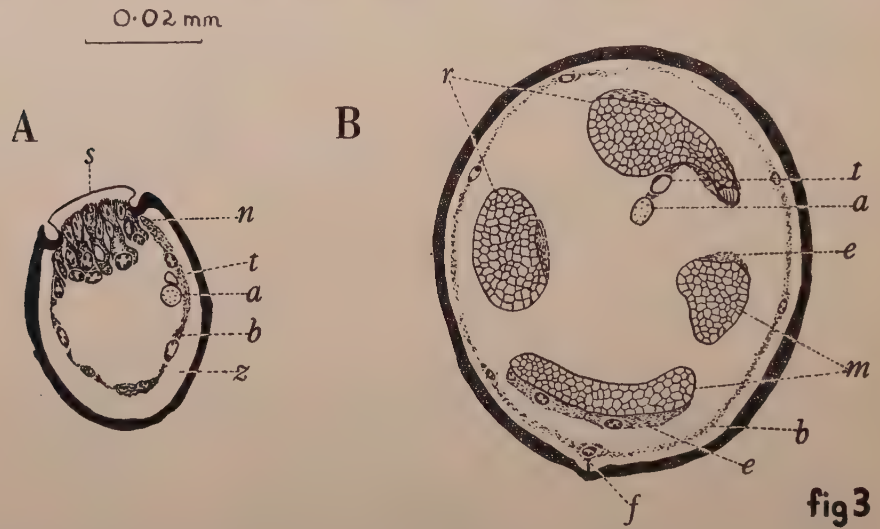

Fig. 3.—Structure of the antenna of *A. fabae* as seen in transverse section. A, 3rd antennal joint showing a secondary sensorium; B, 1st joint (semi-diagrammatic). a = antennary nerve, b = hypodermis, e = end plate of motor nerve fibre on muscle, f = nerve fibre to base of hair, m = extensor muscles, n = sense cells, r = retractor muscles, s = sense plate, t = trachea, z = space due to shrinkage of tissues in fixation.

antennal muscles ending in distinct cells or end plates on these muscles. Similarly in *A. fabae*, no separate motor nerve was found, and the branches and end plates on the muscles agree with those of *P. bursarius* as figured by Pflugfelder (fig. 3, B). Each hair is innervated by a bipolar nerve cell, which links up with the antennary nerve. The trichogen cell (Snodgrass 1926) is generally visible near the sense cell (fig. 2, E). The structure of the primary and secondary sense plates is essentially the same (fig. 2, B, C, D). In both, a group of sense cells lies beneath a thin cuticular membrane and each cell connects proximally with the sensory nerve by means of a fine nerve fibre. There does not appear to be a space behind the membrane, although the cellular matter often tends to shrink away from the cuticle during fixing. The sensoria of *Aphis platanoides* as figured by Flögel (1905) are very similar. In the 2nd antennal joint is a well-developed Johnston's organ (fig. 2, A) (Eggers 1923).

The sense cells are connected to the thin membrane attaching the base of the 3rd joint to the distal end of the 2nd. Proximally the cells link up with the antennary nerve. Attached to the 1st antennal joint are two muscles, an elevator and a depressor, which have their origin on an apodeme of the head. Four muscles, a pair of extensors and a pair of flexors, are inserted on to the base of the 2nd joint, and are similar in arrangement to those found by Miller (1933) in the Woolly Aphis of Elm. There is a trachea running throughout the length of the antenna, often near the nerve. It is most readily seen in an excised antenna from a living aphis mounted in 70% alcohol, and gives off fine tracheoles to the living cells of the antenna.

The internal structure of the apterous forms is very similar, but the sense cells of the secondary sensoria are lacking.

Fig. 5 shows the antennal structure of *Melanoxantherium salicis*. The secondary sensoria, which are relatively smaller than those of *A. fabae*, also consist of groups of nerve cells behind a thin chitinous plate, but there is a tendency for the nerve fibres to run backwards parallel to the antennary nerve for some distance before actually joining it, so that the nerve may appear double in some transverse sections. Groups of nerve fibres from the cells of Johnston's organ run back separately as far as the middle of the 1st joint (fig. 5, A). The antennary nerve was traced back into the brain, where it entered the deutocerebrum. What appears to be coagulated blood is often visible in the centre of the antenna, especially in joints 5 and 6 (fig. 5, C & D).

The internal structure of the antenna of *Macrosiphum pisi* is very similar. The cuticle is thicker everywhere than in *A. fabae.* Blood cells and connecting protoplasmic strands are present in the centre, often around the nerve and trachea. Darkly staining blood is especially well marked in the terminal joints.

fig 4

FIG. 4.—Reconstructed longitudinal section showing internal structure of antenna of *A. fabae*. a = antennary nerve, b = hypodermis, c = cuticular hair, d = depressor muscle, e = end plate, f = nerve fibre, h = hair, j = Johnston's organ, l = elevator muscle, m = extensor muscle, n = sense cell, p = primary sensorium, r = retractor muscle, ss = secondary sensorium. Average length of antenna = 1·47 mm. (0·06 : 0·07 : 0·35 : 0·26 : 0·24 : 0·49 mm.).

The antenna of *Myzus persicae* is almost identical in structure with that of *A. fabae.*

From the simplicity of the structure of the sense organs on the antennae of the four aphids examined, complex responses to external stimuli such as those

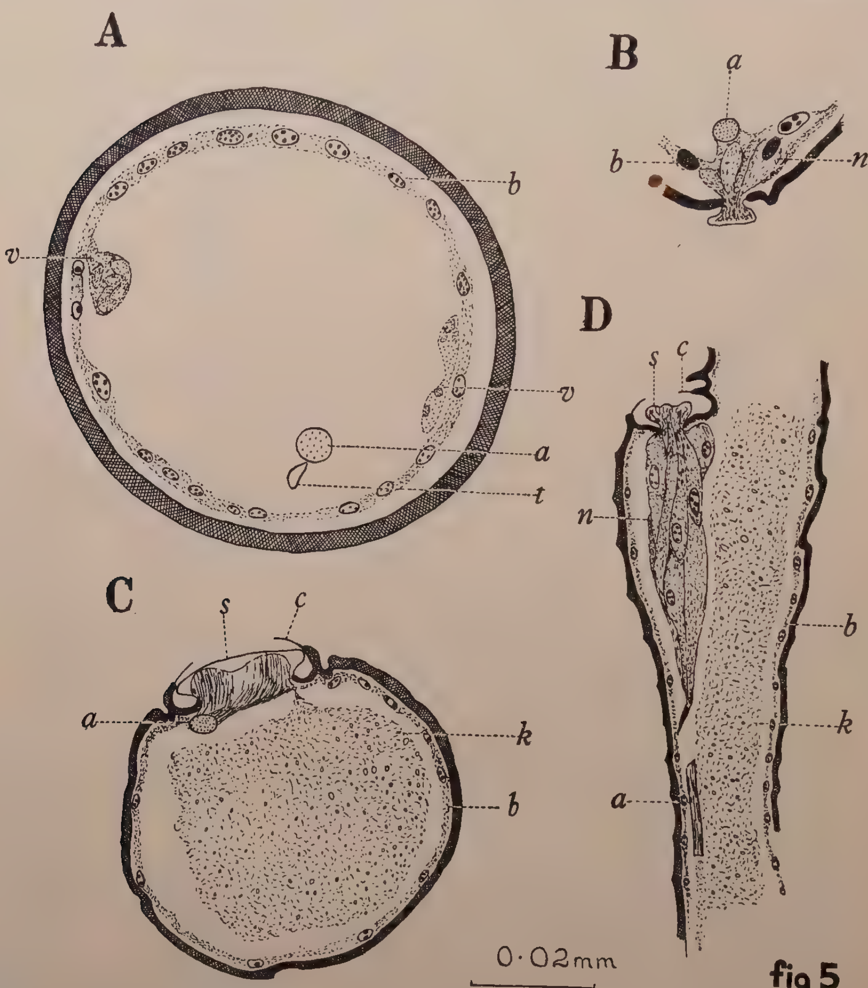

FIG. 5.—Structure of the antenna of *Melanoxantherium salicis*. A, transverse section of 2nd joint, proximal to Johnston's organ; B, transverse section of 3rd joint to show secondary sensorium; C, transverse section of 5th joint, through top of the primary sensorium; D, longitudinal section of base of 6th joint, through a primary sensorium. a = antennary nerve, b = hypodermis, c = cuticular hair, k = blood, n = sense cell, s = sense plate, t = trachea, v = sensory fibres from Johnston's organ.

found in the honey bee (v. Frisch 1919; Minnich 1932) and in the higher Diptera are unlikely.

Work on other insects has shown that sense plates can be regarded as chemoreceptive, while hairs are tactile and probably also chemoreceptive by nature of the thin hair membrane.

As it would be useful to know whether the alatae viviparae of *A. fabae* are able to distinguish plant odours, preliminary experiments were carried out, using an olfactometer.

2. The response of the alatae viviparae of *A. fabae* to external stimuli.

Method.

A slightly modified version of the y-shaped olfactometer (Thorpe and Jones 1937, Thorpe 1938) was used to test the responses of the alatae viviparae of *A. fabae* to host-plant odours.

A current of air was drawn through a bottle containing plant extract on one side and through distilled water or through another plant extract on the other. In some cases, the plant extract was replaced by a bottle containing leaves, and the distilled water by a damp bottle. The flow of air was regulated by means of flowmeters. It was found necessary to have the apparatus running for half an hour before introducing the Aphids, in order to ensure that it was working properly. The apparatus was tested by introducing alatae when the air currents were passed through bottles of distilled water only, and during the experiments by reversing the positions of the bottles periodically. No bias towards any particular side was ever established. The experiments were carried out at a temperature of 25° C. and at a room humidity of 70%. The alatae were reared on Spindle tree (*Euonymus europaeus*), Dock (*Rumex obtusifolius*) and Bean (*Vicia faba*), in muslin cages, and also collected in the field. After collection they were kept in the constant temperature room for at least half an hour before being introduced into the apparatus. The trials were carried out in May, June, and early July, using spring and summer migrating alatae. The results are given in Tables 2 and 3.

Where the difference in response from 50% does not exceed twice the standard error, there is no significant deviation from a chance distribution. In only one case is there any significant deviation from a 50 : 50 distribution. This one positive result to spindle odour was by progeny descended from one

Table 2.

The results of the olfactometer experiments using one plant odour.

Plant extract used	Aphids bred on	No. of trials	Total no. of Aphids	No. to plant	No. to other side	S.E.	χ^2	Probability
Spindle extract [1]	Spindle	9	112	73 65%	39 35%	4·7%	3·56	0·90–0·80
Spindle extract [2]	Spindle	5	288	144 50%	144 50%	2·95%	13·85	0·02
Bean extract .	Spindle	8	193	95 49·21%	98 50·79%	3·6%	4·77	0·70–0·50
Bean extract .	Dock	3	71	38 53·5%	33 46·5%	5·93%	0·53	0·80–0·70
Bean extract [2] .	Bean	3	64	38 59·3%	26 40·7%	6·25%	6·74 (59 : 41 basis)	0·05–0·02
Dock extract .	Dock	4	179	82 48·5%	87 51·5%	3·8%	0·896	0·90–0·80
Bean leaves .	Bean	3	99	51 51·5%	48 48·5%	5·02%	2·69	0·30–0·20
Bean leaves .	Spindle	6	295	147 49·8%	148 50·2%	2·90%	10·81	0·10–0·05

[1] Alatae bred from one fundatrix.
[2] Not homogeneous.

S.E. $= \frac{pq}{n}$ where $p = 0{\cdot}5$, $q = 1{-}0{\cdot}5$, $n =$ number of Aphids. For significance, deviation from a 50 : 50 distribution should equal 2 × S.E. Tables given in Fisher (1938) were used to look up Probability.

TABLE 3.

The results of the olfactometer experiments using two plant odours.

Plant used	Aphids bred on	No. of trials	Total no. of Aphids	No. to plant 1	No. to plant 2	S.E.	χ^2	Probability
Spindle extract = 1 Dock extract = 2	Spindle	3	72	34 47·2%	38 52·8%	5·89%	0·61	0·80–0·70
Bean extract = 1 Dock extract = 2	Dock	4	307	156 50·7%	151 59·3%	2·85%	1·77	0·70–0·50
Bean extract = 1 [3] Dock extract = 2	Bean	3	273	151 55·3%	122 45·7%	3·03%	6·13 (55 : 45 basis)	0·05–0·02
Bean extract = 1 Dock extract = 2	Spindle	1	44	22 50%	22 50%			

[3] Not homogeneous.

fundatrix, and not by a mixed population. The homogeneity of the readings as calculated by χ^2 was satisfactory in all trials except three. In these three trials, one in which alatae bred on spindle were tested with spindle extract and two in which alatae bred on bean were tested with bean extract, a significant positive response is nearly achieved, and further work is necessary before any definite conclusion can be drawn. These two plants are the chief winter and summer hosts of the Aphid.

The results, therefore, so far as they go, do not indicate any well-marked olfactory sense.

Davies (1935) showed that high humidities have an inhibitive action on the flight of *Myzus persicae* Sulz., and also that differing humidities probably have an effect on the aphid population on potatoes in North Wales. This is presumably due to the effect of humidity on flight. The reaction of *A. fabae* to two air currents of differing humidities was tested in an olfactometer. The bottles containing plant extract were replaced by four bottles, two in series containing calcium chloride and two in series containing glass beads in distilled water, so that in one case the current was almost saturated and in the other almost dry. The experiments were, as before, carried out in a room with a relative humidity of 70%. The results are given in Table 4, and show a significant reaction away from the damp current. A calculation of χ^2 shows that the trials were homogeneous.

TABLE 4.

The results of olfactometer experiments using currents of differing humidities.

Number of trials	Number of Aphids used	Number of Aphids to dry side	Number of Aphids to wet side	S.E.	χ^2	Probability
10	578	336 58·2%	242 41·8%	2·08%	8·52	0·70–0·50

3. Discussion.

The alatae viviparae of *A. fabae* serve chiefly to distribute the species to new host plants over a wide area, and it is significant that they bear sensoria on the 3rd and 4th antennal joints, while the apterae, which remain on one host plant or migrate by walking to neighbouring host plants, have hairs only on these

segments. Detailed examination of the body surface of both alatae and apterae shows few types of sense organs. Two compound eyes are found on the head, while at the end of the labium are a group of small tactile hairs, which, according to Davidson (1914), enable the Aphid to find a suitable tender part of the plant into which to insert its stylets. Hairs are present on all segments and on the limbs. Davidson (1914) showed that there is a gustatory organ in the roof of the mouth in *Schizoneura lanigera* Hausmann, which would appear to appreciate different plant juices. This organ is probably present in *A. fabae*. Thus, so far as sense organs are concerned, the alata is distinguished from the aptera only by the presence of secondary sensoria on the antennae and a small median ocellus on the head. It seems very probable, therefore, that the sensoria on the antennae are connected with flight. The oviparous female, as an adaptation in connection with the egg-laying habit, does possess a number of simple sense plates on the hind tibiae, where the number of hairs compared with other forms is small (Jones 1942).

Olfactometer experiments do not demonstrate any well-marked response to host-plant odours, although there are indications that some colonies may be more responsive than others. When currents of differing humidities are passed through the apparatus, the response of the alatae to the drier side is definite, but not great. Davies (1936) found that the velocity of the wind has a marked effect on the flight of *Myzus persicae*. Above a velocity of 3·75 m.p.h. flight ceased and at higher velocities the alatae remained adpressed to the walls of the apparatus used. Field observations on *A. fabae* confirm these findings, for on a warm, dry, calm day more alatae are on the wing than on a windy or wet day. The velocity of the air currents used in the above experiments, unfortunately, was not measured, but the presence of flowmeters ensured that it was the same on both sides. In every case, the alatae walked towards the light, but when the current was turned off, many flew into the traps at the ends of the arms. It seems very likely, therefore, that one of the functions of the sensoria is to indicate the suitability of humidity, and perhaps wind velocity, for flight.

The increase in the number of sensoria on the antennae of the autumn migrants (Table 1) which return to a restricted number of winter hosts (*Euonymus europaeus* and *Viburnum opulus*) as compared with the wide range of summer hosts is interesting, and may allow a greater response to external stimuli. The further increase in the number of sensoria on the antennae of the males may be correlated with the more difficult task of finding the oviparae.

4. Summary.

1. The external and internal structure of the antennae of the alatae viviparae of *Aphis (Doralis) fabae* Scopoli are described. One antennary nerve is present, and the sensoria and hairs are innervated by well-marked sense cells.

2. The internal structure of the antennae of the alatae viviparae of *Melanoxantherium salicis* L. shows only very small differences from that of *A. fabae.*

3. The essential similarity of structure of the antennae of the alatae viviparae of *Macrosiphum pisi* Kaltenbach and *Myzus persicae* Sulz. with that of *A. fabae* is noted.

4. When a population of alatae viviparae of *A. fabae* of mixed origin is passed through an olfactometer, there is no well-marked response to the odour of host-plant extracts or leaves, but there is a small, positive response to a dry current as compared with a wet one.

5. One of the functions of the secondary sensoria on the antennae of the

alatae viviparae may be to indicate the suitability of weather conditions such as humidity and wind velocity, for flight.

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SIMPLE EXPERIMENTS ON THE BEHAVIOUR OF BODY LICE (SIPHUNCULATA)

By J. R. BUSVINE, B.Sc., Ph.D.

(*Ministry of Health Entomologist.*)

ALL the experiments to be described consist of the release of lice on or near to the human body and the search for them after an interval of time. The results show where and how far lice are likely to travel under certain circumstances and provide some information as to how readily a man may become infested.

Pioneer experiments of this kind were described by Lloyd (1919), who showed how readily lice will pass from one man to another sleeping in the same bed, especially if the primary host is in a febrile condition.

SERIES 1. NATURAL WANDERING AMONG UNDERGARMENTS.

Away from the body, and perhaps impelled by the desire to find a new host, the louse moves at a considerable speed : Buxton (1939) records 9 inches in a minute at 20° C. But, unless it is driven in one direction by unilateral lighting, the insect meanders about so that the distance covered from point to point is much less than would be expected.

There seems to be no recorded information about wandering under natural conditions of infestation, *i.e.* under the clothing. If one examines naturally infested underwear immediately after removal from the body, the insects do

not appear to be very active unless the garments are taken near to a window, when they will walk away from the light. A high proportion of the lice seem to have made a comfortable niche for themselves against one of the seams of the garments. Nevertheless, the presence of lice and eggs on outer garments proves that the insects must sometimes move away from their source of food, the skin.

The lice used in all the tests to be described were taken from a stock which had been reared in pill boxes for many generations. There is no reason to suppose that they would tend to wander more than " wild " lice.

Method A. A dozen adult lice were placed at a spot inside a fleecy woollen vest which lies against the skin of the small of the back when the vest is worn. The vest was put on, together with aertex short pants, a poplin shirt, flannel trousers and a laboratory overall. Three or four preliminary trials proved that putting on the vest did not dislodge the lice.

After two hours (during which the host mainly sat quietly) the garments were all quickly removed and a search made at once for the lice.

The figures in Table 1 give the *minimum* distances travelled during the experiments : the average is about a foot. In many cases the lice walked from the inner to the outer surface of the vest, and sometimes as far as the outside of the shirt. In the terms of MacLeod and Crauford-Benson, they had migrated from one " territory " to another.

It will be seen that the pubic hair was infested in three of the four tests and eggs were found to have been laid on this site. It is clear that body hair must not be overlooked in de-lousing operations.

Table 1.

Location of lice found 2 hrs. after release at the small of the back. Figures in brackets are minimum distances wandered in inches.

Test No.	Lice found	Lost	Average minimum distance, inches
1	2 on pants (12) (16), 2 inside shirt (25) (10), 3 on vest (10) (22) (6), 3 on pubic hair (15) (15) (15).	2	14·6
2	1 on pants (10), 1 on shirt (16), 7 on vest (18) (16) (16) (16) (16) (6) (3), 2 on pubic hair (15) (15).	1	12·0
3	1 on pants (18), 5 on vest (18) (12) (6) (6) (2), 3 on pubic hair (15) (15) (15).	3	12·0
4	3 on pants (16) (16) (10), 6 on vest (14) (8) (8) (8) (8) (1).	3	9·2

In these experiments the lice were liberated on uninfested under-garments; the results therefore only represent the start of an infestation, not an established case. Wigglesworth (1941) has shown that lice react favourably to the smell of other lice and to their excrement and suggests that this partly explains their aggregation at the seams of clothing. However, the following test indicates that even this attraction does not inhibit a great deal of wandering of the adults.

Sixty lice were confined beneath a three-inch square of bolting silk on the inside back of a vest. Knotted cord sewn to the vest gave them room to feed and provided a convenient resting place. After three days the bolting silk and lice were removed, leaving a contaminated patch round the knotted cord.

A liberation experiment was performed as above with a dozen adults from the box cultures. Two hours after they had been carefully placed on the knotted cord and the vest worn, they were found as follows : 1 outside pants (25), 2 on shirt (18 & 18), 6 on vest (0, 8, 10, 11, 12, 15), 1 on pubic hair, 2 lost. Average minimum distance 13 in.

Method B. These tests were similar to the foregoing except that the lice were released on a pleated cotton belt which discouraged wandering (because lice tend to settle down in folds and pleats). Nymphal as well as adult lice were used and in two cases the host carried out half an hour's physical exercise in thick clothing during the course of the experiment to promote sweating.

Ten adult and 10 second- or third-stage nymphs were released in each test, the results of which are shown in Table 2.

TABLE 2.

Amount of wandering from a cotton belt with and without exercise and sweating.

Test No.	Conditions.	Adults			Nymphs		
		No. wandered	Average distance wandered, inches	No. lost	No. wandered	Average distance wandered, inches	No. lost
1	Sitting quietly	5	4·0	3	2	?	2
2	2 hours	6	6·6	0	2	1·2	0
3	Exercise ½ hour	9	10·3	2	6	5·4	1
4	Sitting 1½ hours	7	9·5	1	3	?	3

It will be seen that the physical exercise and sweating tended to increase the wandering of the lice. This had been suspected from observations on naturally infested men, and it is not a very surprising fact, for the increase in skin temperature would naturally increase the activity of the lice.

In all cases the adults wandered much farther than the nymphs. This also is in harmony with observations on infested men by MacLeod and Craufurd-Benson (1941b), who found wandering lice on the outer clothing to consist of proportionately more adults than the populations on garments next the skin. It seems probable that new infestations are caused mainly by wandering adults.

SERIES 2. ACQUIRING LICE FROM A LOUSY BED.

Method : A cotton sleeping-bag was used measuring 6 ft. × 2½ ft. About 20 adult lice were put into the bag in the morning and the bag was rolled up and left with the opening directed towards a window to lessen the chances of escape (nevertheless, in some tests, several did escape during the course of the day !). In the evening the lice present were counted and a man wearing pyjamas slept in the bag during the ensuing night. The sleeping-bag was covered by two woollen blankets and a light eiderdown, as the weather was rather cold.

Next morning a careful search was made for the lice. The results are recorded in Table 3.

As might be expected, lice were more often picked up by the fleecy surface of the flannel than the smooth surface of the poplin pyjamas. It is interesting

TABLE 3.

Distribution of lice found after sleeping in an infested sleeping-bag.

Test No.	No. lice in bag in evening	Pyjamas worn	Percentage lice next morning				
			On pyjamas	On body	Inside bag	Outside bag	Lost
1	16	Flannel	50	6	6	13	25
2	20	Poplin	10	5	45	15	25
3	20	Flannel	40	5	20	15	20
4	20	Poplin	15	0	45	40	0

to note that 35% to 40% escaped during the night, which meant travelling a *minimum* distance of 6 ft. These lice often became entangled in the blankets in which they could only be discovered by a very careful search. This migrating habit of the lice seems to be the explanation of the observation of MacLeod and Craufurd-Benson (1941a), who remarked that casual beds become un-infested a few days after lousy individuals have slept in them.

SERIES 3. TESTS WITH AN ANTI-LOUSE TYPHUS PROTECTION GOWN.

A protective gown for medical and sanitary personnel (who may come in contact with typhus-infested lice) is described in the Ministry of Health Memorandum on louse-borne typhus fever 252/med. (1941). This garment gives complete protection except for the openings at the back, round the face and at the wrists. A zipp fastener is recommended for sealing the large opening at the back, but these fasteners do not well endure the continual sterilisations necessary. As an alternative, tapes are suggested, to be tied over an 8-in. overlap. Dr. Cosins, Medical Superintendent, Orsett Lodge Hospital (Essex C.C.), has proposed the following modification of the gown and suggested that insecticide treatment might be used in some way to enhance protective value. The two edges of the back opening are extended to form a flap which can be rolled up and the tapes tied across it. Round the face is introduced a double edging of gauze between which is a cotton-wool pad. A gown altered in this way was submitted to test with and without the application of Lethane 384, an effective anti-louse insecticide.

Method. The gown was put on by a man wearing ordinary clothes but no jacket and fastened in the prescribed manner. A known number of body lice were placed on the outside of the garment near the face opening and the rolled flap at the back. The wearer sat or walked about in a laboratory in daylight, not directly in the sun. After an hour a careful search was made to find if the lice had penetrated to the inner surface.

The protective effect of Lethane 384 was tested by lightly spraying the cotton gauze round the face opening and the edges of the back flap : about 2–4 c.c. of undiluted Lethane were used.

Results. Lice which crawled into the rolled flap were not considered to have penetrated the gown unless they passed right on to the *inner surface* of the two rolled edges. Their power of penetrating into recesses of garments is really extraordinary : in the first test one louse had found its way past the rolled edges, under the wearer's shirt, and as far as the armpit.

TABLE 4.

Intrusion of lice into protective clothing.

Test No.	Remarks	No. of lice released	Percentage of lice		
			Found outside	Found inside gown	Lost
1	No Lethane	36	72	14	14
4	No Lethane	24	46	17	37
2	Lethane used	22	91	0	9
3	Lethane used	24	67	0	33
5	Lethane used	24	71	0	29
6	Lethane used	24	88	0	12

Each test is fairly severe, for the attack of so many lice in the vicinity of the openings would correspond to many weeks' wear. Nevertheless it is rather alarming to find that a few lice were able to penetrate the untreated garment. The Lethane application gave complete protection as far as these tests went. This is because the lice which crawled into the vicinity of the two main openings were found afterwards to be stupefied by the insecticide and subsequently died.

It is evident that spraying the openings of anti-louse typhus gowns with Lethane will considerably increase their protective value. The spraying can be done with any small hand sprayer such as a " Flit " gun, a scent spray or an artist's pastel-fixing spray.

The insecticide is removed by laundering or heat sterilisation and should be applied every day immediately before putting on the garment. As only 2–4 cubic centimetres are used per application, a U.K. gallon of the Lethane 384 will suffice for one or two thousand treatments.

The insecticide is liable to cause stinging if it touches the face. Care was therefore taken not to spray the *inside* face gauze.

ACKNOWLEDGMENT.

I would like to record my appreciation of the help and encouragement of Professor P. A. Buxton, F.R.S., who suggested this type of experiment and carried out a preliminary test on himself.

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EVIDENCE OF EXTENSIVE DESTRUCTION OF *DANAUS CHRYSIPPUS* L. (LEPIDOPTERA) IN S.W. ABYSSINIA

By Professor G. D. Hale Carpenter, M.B.E., D.M., F.R.E.S.

With a Note by Lt.-Col. F. C. Fraser, M.D., I.M.S. Rtd., on Method of Capture by Mantids.

Major T. H. E. Jackson wrote in February, 1942 : " A large bush of *Capparis* was found in full flower in South-West Abyssinia in January, just after the short rains, down on the Omo river, at about 1500 feet elevation just twenty miles north of where it enters the Lake Rudolf. The bush was attracting immense numbers of butterflies, chiefly common *Colotis* and *Belenois severina* and *infida* with a few *Precis* and a few more *Danaus chrysippus*. Suddenly I noticed the ground covered with wings, so with the help of my three or four Turkhana constabulary we collected them all. The result is surprising. Although I watched carefully I could not find the culprit." Replying to a query Major Jackson wrote later : " I looked hard for mantids and although I did not find any I feel sure they were to blame." The remarkable point about the collection of wings, and damaged butterflies, is that almost all are *D. chrysippus* in one form or another. Anyone who has seen *Capparis* bushes in flower knows how various species of *Colotis* throng to the flowers and oviposit on the bushes. Yet among the wings collected on the ground there were remains of only 4 specimens of *Colotis*, 2 of *Belenois*, 1 *Danaus limniace petiverana* Suff., 1 *Dixeia*, 1 *Acraea* probably *neobule* Dbl., 1 *Precis oenone cebrene* Tr. and 1 Sphingid : all the rest belonged to *D. chrysippus*, representing 104 specimens at least. This points to selection of prey, for in such environment species of *Colotis* normally far outnumber the *Danaus*. The data given below show also the proportion of females to be not that familiar to collectors, for it is far higher. The great excess of the form *dorippus* Klug is characteristic of the locality.

I am indebted to my senior laboratory assistant, Mr. E. Taylor, for putting together the following details of the five forms represented.

f. *aegyptius* Schreber. R.F.W., 9. L.F.W., 4.
f. *transiens* Suffert. R.F.W., 2. L.F.W., 1.
f. *dorippus* Klug. ♂ 6, ♀ 8, of which 4 ♂, 1 ♀ lack R.F.W., 2 ♀ lack L.F.W., and 1 ♀ lacks both F.W. Also 55 R.F.W., 74 L.F.W., 12 ♂ R.H.W., 18 ♂ L.H.W., 16 ♀ R.H.W., 20 ♀ L.H.W.
f. *albinus* Lanz. ♂ 1. 1 ♀ L.H.W.
f. *semialbinus* Strand, ♂ 2 (one lacking L.F.W.). 1 ♀ L.H.W.

An estimate of the total number of specimens destroyed can only be approximate, as allowance must be made for the possibility that some at least of the isolated wings might belong to dismembered specimens.

But taking the following figures a total of 104 butterflies destroyed can be deduced. 9 R.F.W. of *aegyptius*, 6 male and 8 female *dorippus* more or less complete, 74 L.F.W. of *dorippus*, 2 R.F.W. of *transiens*, 1 whole male *albinus* and 1 female L.H.W., 2 whole male *semialbinus* and 1 female L.H.W.

The possible predators are birds and MANTIDAE. Major Jackson saw no mantid on the bush, and a bird which might have been the predator would have flown away on the approach of the party before the wings were noticed. I am, however, of the opinion that it was the work of a bird. Firstly, on account of the evidence of selection of the female sex against the male. Secondly, because there is not the scratching of the wings to the degree which might be expected from a butterfly held fluttering while the body is eaten. Thirdly, because there are on six of the specimens clear beak-marks *all of the same size and shape.* It would indeed be a strange coincidence if the only butterflies, bearing previously inflicted beak-marks, which were devoured by a mantid, should all have been attacked by the same kind of bird. The beak-mark is small and of the type figured as L—O on Pl. 1 of 1933, *Trans. R. ent. Soc. Lond.* **81**. The *Proceedings* of the Society for 1937, (A) **12**: 161–162, contains an account of a litter of wings found under a nest of a shrike, which was seen " pulling a butterfly to pieces ".

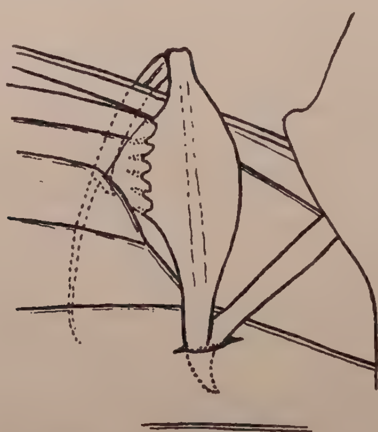

FIG. 1.—The suggested formation of a false beak-mark by the fore leg of a mantid.

If the predator in Abyssinia was a bird the case is analogous to the oft-quoted *Artamus fuscus* (Ashy Wood-swallow) noted by Fryer in Ceylon to hawk systematically *Euploea* and *Danaus.*

Since communicating this note I have received the following interesting letter from Lieutenant-Colonel F. C. Fraser, M.D., I.M.S., dated 11th October, 1943. " I was much interested in the note you read before the R. ent. Soc. meeting lately, of numbers of butterfly wings being found beneath a bush of Caper. It reminded me that I had seen this phenomenon on numerous occasions, and, by a curious coincidence, nearly always beneath Capers. In all these cases the wings were the ' hang-over ' of the larder of a praying mantid (*Gongylus gongylodes* L. usually), so that in the search for these insects I came to look for the carpet of wings as evidence of their presence. The prothoracic flaps or processes of the mantid are pale peach-blossom in tint whilst in the living state, and are held up to view in the usual attitude adopted by the insect, so that passing butterflies are apt to mistake it for a blossom amongst the otherwise arid nakedness of Capers. They approach and are mowed down by a sweep of the mantid's fore legs, the wings being transfixed by the long spines. I think it would be of interest to examine the wings for evidence of such impalement. The mantid having locked the butterfly securely in the rat-trap-like fore limbs

then rapidly bites off the wings at the base. In so far as I can remember the majority of the wings were Pierid or Papilionid (*Colotis*, *P. hector* L. and *P. aristolochiae* F.) but I seem to remember *D. chrysippus* L. also fell to the snare."

According to Col. Fraser's suggestion, I have re-examined the specimens exhibited, and find that many do show perforations of the disc of the wing, sometimes like a pin-hole, sometimes a jagged tear. This increases the evidence for the predator being a mantid, but it is unusual, I believe, for these insects to show selective preference, which has certainly happened in this case.

Note by Lt.-Col. F. C. Fraser: Professor Carpenter is of opinion that there would be some scratching of the wings caused by the fluttering of the butterfly whilst being held and eaten, but there never is any fluttering: from the initial stroke made by the mantid, its victim is literally smothered, in so far as any movement is concerned, and there is not so much as a kick out of it.

With regard to the "selective preference," may this not be more apparent than real? May not the *Danaus* be more open to attack than the Pierids? *Danaus* alights with a lazy flutter: its wings are held more or less open and thus offer an easy target for the downward, scimitar-like stroke of the mantid's fore legs: probably a hundred per cent. of these attacks are successful. Pierids alight with great directness and close their wings with a snap, and so the downward stroke of the mantid is apt to sheer through the air harmlessly on either side of them. All now depends on their quickness in making a get-away before the mantid can get in another stroke; it is probable that the majority which fall to the snare are caught by this latter as they open their wings to make off, but a large number might escape the first attack. The mantid makes its stroke with the legs held somewhat divergent so that the elbow-spine transfixes the wings at about the discal area: thus a butterfly alighting with its wings closed would be much more likely to escape impalement than one with its wings held open. After transfixing the wings, the tarsus is passed over the costal border of the victim's fore-wings, and the wings thereon are securely clamped in the apposed spiny borders of the tibia and tarsus. It is even possible that this inclusion of the wings in the V formed by the strongly flexed tarsus on the tibia might at times cause a false beak-mark to be imprinted on the wings.[1] If so, then this marking would always be on the fore-wing with the apex of the V towards the costa, and would be margined by the fine punctures of the tibial and tarsal spines.

The absence of Major Jackson's mantid is sufficiently explained by the enormous size of its larder, which points to it being a very old adult, which had either died or moved off to deposit its eggs. Never more than one female is found on any one bush: they are quite inimical to one another, so that the whole of these butterflies must have fallen victim to a single mantid.

The figure reproduced (Fig. 1) shows two possible sites for false beak-marks, produced by pressure of combined femur and tibia, or tibia and tarsus. The perforation of the wing by the elbow-spur and the grip across the costal border are also demonstrated.

[1] The marks made according to Lt.-Col. Fraser's suggestion would have little in common with beak-marks, for they would lack the apical angle and would only be shown on *one* surface of the wing.—G. D. H. C.

STAGES IN THE RECOGNITION OF BIOTIN AS A GROWTH FACTOR FOR INSECTS

By G. Fraenkel and M. Blewett.

(Department of Zoology and Applied Entomology, Imperial College of Science and Technology, London.)

We have shown elsewhere (Fraenkel and Blewett 1943a) for several insects that a diet consisting of casein, glucose, cholesterol, salt mixture, and yeast is very much superior to the same diet in which yeast is replaced by aqueous yeast extract. These results were obtained with five different beetles: *Tribolium confusum* Duv., *Sitodrepa panicea* L., *Lasioderma serricorne* Fab., *Silvanus surinamensis* L. and *Ptinus tectus* Boield. The need of insects for a water-insoluble factor in yeast has been shown before by Hobson (1933) for *Lucilia*, Fröbrich (1939) for *Tribolium*, and Tatum (1939, 1941) for *Drosophila*, and seems therefore of widespread occurrence. In searching for this "insoluble factor" certain similarities with vitamin H gradually emerged, and after vitamin H and biotin had been found to be identical by Du Vigneaud *et al.* (1940), and pure biotin became available, it was possible to prove that the "insoluble factor" necessary for insects and biotin were identical. Short accounts of some of the results have appeared elsewhere (Fraenkel and Blewett 1942a & b). This paper contains the steps which led to the recognition of biotin as a growth factor for insects.

Experiments.

The technique of experimenting and assessing the results is described elsewhere (Fraenkel and Blewett 1943a & b). The curves which demonstrate the relative deficiency of diets containing yeast extract in place of whole yeast are given in Fraenkel and Blewett (1943a, figs. 7–11). The following diet was used: casein 50, glucose 50, cholesterol 1, McCollums salt mixture 1, and yeast extract (prepared according to Chick and Roscoe 1930) 15 parts. This diet will be referred to, in this paper, as the "yeast-extract diet." On this, *Tribolium* pupates usually after about 40 to 45 days. But on some occasions growth has been slower than that (fig. 2), and on others quicker. The difference in the efficiency of this diet, encountered with different preparations of yeast extract, and on different occasions, is almost certainly mainly due to different amounts of traces of the insoluble factor. This is shown in a test in which the concentration of the yeast extract had been trebled, and in which the efficiency of the diet was greatly improved (fig. 1). Therefore comparisons between the efficiency of different diets can only be made within a batch of simultaneous tests, containing the same sample of yeast extract.

The yeast-extract diet is also considerably improved by adding Marmite (fig. 1). Marmite is autolysed yeast and would, therefore, be expected to contain the insoluble factor, unless it were destroyed in the course of its manufacture.

A liver extract, supplied by Glaxo Laboratories, proved to be inferior to a yeast extract (fig. 2). The factor missing in the liver extract seems to be the same as the factor missing in yeast extract. The combinations, liver extract

plus insoluble liver, liver extract plus insoluble yeast, and yeast extract plus insoluble liver, are of similar efficiency to yeast extract plus insoluble yeast.

Casein is a protein which is chemically very different from the proteins of wheat, a natural food for all the insects under investigation. A yeast-extract diet might be deficient in some protein constituent (amino-acid) which would be supplied by yeast or insoluble yeast. If so, one might expect that gliadin, a wheat protein with a chemical composition very different from that of casein, would supply amino-acids missing in casein and so improve the yeast-extract diet. But using 20 parts of gliadin plus 30 parts of casein, instead of 50 parts of casein, had the effect of making the diet noticeably inferior (fig. 2). Toxic effects of large amounts of gliadin in the diet of rats have been described by Melnick and Cowgill (1937).

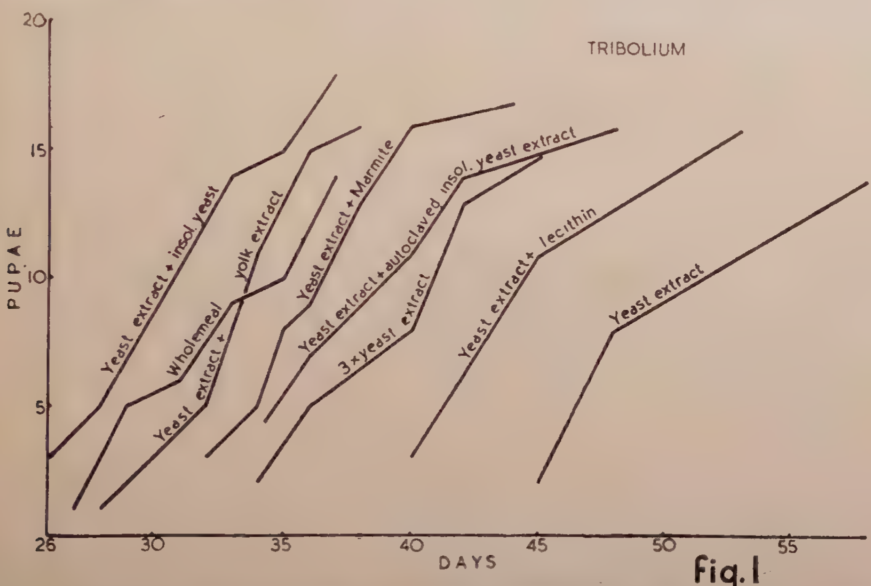

FIG. 1.—Growth of *Tribolium confusum* on the "yeast-extract diet," consisting of casein, glucose, cholesterol, salts and aqueous yeast extract, to which various substances or fractions have been added.

Insoluble yeast contains all, or much, of the fat soluble substances of yeast, and it was considered possible that the insoluble factor might be fat-soluble. However, extracting insoluble yeast with petrol ether for 10 hours does not in the least alter its activity in supplying the insoluble factor. In another test, insoluble yeast was extracted with chloroform, and the extract improved the yeast-extract diet only very slightly. If the insoluble factor were fat-soluble it would be expected that wheat germ oil contained it. Wheat germ oil somewhat improved the yeast-extract diet (fig. 2). When the oil was divided into the saponifiable and unsaponifiable fractions, the unsaponifiable parts had the same activity as the oil (fig. 2). From this it would appear that in addition to cholesterol, already in the diet, another unsaponifiable substance may be required by *Tribolium*. This, however, has not been confirmed in later tests. The wheat germ oil and the unsaponifiable fraction were added in the diet in quantities far exceeding the amount present in whole meal flour and yet the improvement induced by these fractions in a yeast-extract diet was only slight.

From this we may conclude that we are here dealing with some additional minor factor, and not with the insoluble factor itself.

These preliminary tests showed that the insoluble factor was contained in the water-insoluble fraction of yeast or liver, that it could be brought into solution to a certain extent by autolysis (Marmite) and that it was probably not fat soluble. These properties suggested a similarity to György's vitamin H, which had at that time just been shown to be identical with biotin, and has been recognised as an essential nutritional factor for the rat (Du Vigneaud *et al.* 1940). Accordingly our efforts were concentrated on finding out whether the insoluble factor was, in fact, biotin. Since pure biotin only became available long after the work had started, it was at first necessary to seek the solution in an indirect way.

Kögl's (1936) original isolation of biotin was made from egg yolk, and he showed in the course of his investigation that commercial lecithin, which is

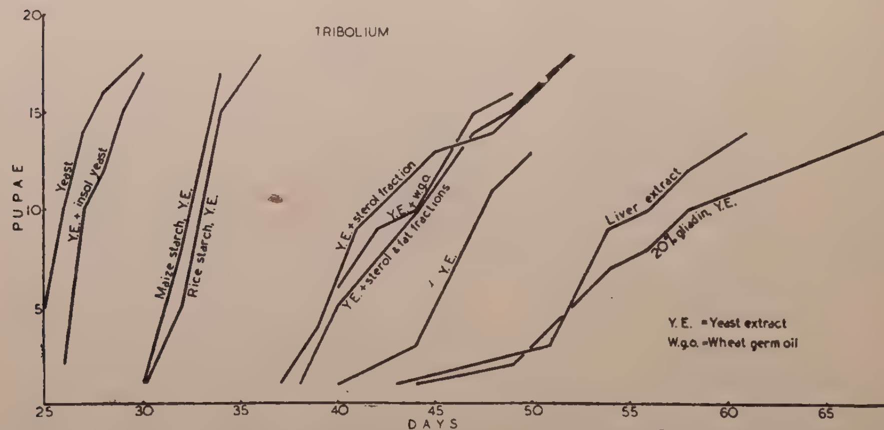

Fig.2

Fig. 2.—*Tribolium confusum.* Explanation as fig. 1. The two curves on the left refer to "yeast-extract diets," in which glucose has been replaced by starch.

made from the same source, usually contains traces of biotin as an impurity. Adding lecithin (B.D.H. comm.) to the yeast-extract diet, in fact, improved it noticeably (fig. 1).

György *et al.* (1939) prepared a vitamin H concentrate from the water-insoluble residue of liver by autoclaving and treating the resultant solution in turn with acetone and alcohol. We applied the same treatment to the insoluble fraction of yeast which was treated according to the following scheme: autoclaved for 5 hours at 130° C., filtered; filtrate treated with 2 parts acetone, filtered; acetone boiled off from filtrate, 2 parts alcohol added and filtered; alcohol boiled off from filtrate. This autoclaved extract from insoluble yeast, added to the yeast-extract diet, improved it very considerably (fig. 1).

The initial steps of Kögl's (1936) original isolation of biotin from yolk were boiling in water and removing in turn all that precipitates in alcohol and acetone. We prepared an extract from the yolk of one egg by the method just described for insoluble yeast, following closely the first steps of Kögl's isolation. The improvement obtained with yolk extract was still very much greater than that obtained with extract from insoluble yeast and brought the

diet close to the efficiency of the controls (wholemeal flour or yeast extract plus insoluble yeast, fig. 1).

Subsequently a sample was obtained of the most powerful biotin concentrate then on the market (Smaco, 200 μg. biotin/ml.). This was fed in a series of concentrations and was found to improve the yeast-extract diet to about the same extent as the yolk extract. The optimum was reached with quantities corresponding to 1 μg. biotin/g. dry food. Experiments undertaken later with pure biotin preparations indicate, however, that the biotin extract might not have been full strength.

The results of tests with autoclaved extract of insoluble yeast, yolk extract and biotin concentrate strongly suggested that the insoluble factor was biotin. This was finally proved with pure biotin methyl ester and pure biotin in experiments which are described elsewhere (Fraenkel and Blewett 1943c).

All the tests so far described in this paper were done with a diet which contained glucose as the carbohydrate. In the course of earlier experiments with *Ptinus*, shown in Fraenkel and Blewett (1943a, fig. 11), it was found that the difference between the efficiency of the yeast-extract diet and the yeast extract + insoluble yeast diet was not as great when starch was used instead of glucose. When *Tribolium* was later reared on a diet with starch instead of glucose, growth was found to be very much better on a yeast-extract diet in the presence of starch than of glucose, and the starch + yeast-extract diet was almost as successful as if it had contained insoluble yeast (fig. 1). Identical results were obtained with two different samples of starch, B.D.H. fat free and vitamin free rice starch, and a specially purified sample of maize starch. It appears from these tests that even purified starch contains sufficient biotin to allow almost optimal growth of *Tribolium*.

Discussion.

Insects require for optimal growth the insoluble fraction of yeast in addition to yeast extract. Hobson (1933) found that the insoluble factor in yeast, required by blowfly larva, could be supplied by Marmite. Fröbrich (1939), working with *Tribolium*, found that growth was poor on a diet consisting of casein, rice starch, arachis oil, ergosterol, salts and yeast extract, and that the deficiency was made good by adding the insoluble part of yeast. On the other hand, the beetle *Silvanus surinamensis* grew well on the same diet without insoluble yeast. From this the conclusion is drawn that "factor U," as the insoluble factor was named, is essential for *Tribolium* but not for *Silvanus*. Our experiments suggest (cf. Fraenkel and Blewett 1943a, fig. 8) that *Silvanus* required biotin, but in smaller amounts than *Tribolium*, and that sufficient biotin may have been supplied in Fröbrich's diet with starch or the yeast extract. Offhaus (1939) grew *Tribolium* on a diet essentially the same as Fröbrich's except that it contained fresh egg white instead of casein. On such a diet, in the presence of yeast extract and a sterol, insoluble yeast was not required. This is easily explained. Egg white is a relatively rich source of biotin, of which it contains according to Lampen *et al.* (1942) 0·53 μg./g. dry weight. Since fresh egg white constituted about 20% of the diet, and roughly one-sixth of egg white is dry matter, the biotin content of Offhaus' diet would be roughly 0·02 μg./g. which is almost enough for optimal growth of *Tribolium*. The need of *Tribolium* for biotin (Fraenkel and Blewett 1942b) has been confirmed by Rosenthal and Reichstein (1942).

In one of the earlier works on insect nutrition Guyénot (1917) stated, that

the *Drosophila* larva required, under sterile conditions, peptone, salts, alcohol-soluble fraction of yeast autolysate and lecithin. It is probable that lecithin was required as a source of biotin. Very recently Tatum (1939, 1941) states that three fractions of yeast are essential for optimal growth of the *Drosophila* larva: Fraction I-water and alcohol insoluble; residue from yeast autolysate. Fraction II-water soluble; alcohol extract of dried brewers yeast. Fraction III-water soluble; precipitated by barium hydroxide and alcohol from yeast extract. Without the insoluble fraction I growth is slow but otherwise normal. This is in agreement with our experience. It is probable that fractions II and III together contained sufficient biotin for slow growth to take place.

Since Kögl (1936) isolated biotin from yolk, it was known to be active in extremely small quantities. This has been confirmed by us. The optimum requirements of *Tribolium* are 0·05–0·1 $\mu g.$/g. dry food, while the threshold of action is lower than 0·006 $\mu g.$/g. (Fraenkel and Blewett 1943c). The biotin content of several foods has recently been determined by microbiological methods and found to be for bakers yeast 0·7, brewers yeast 0·83, egg white 0·53, yolk 1·0, wheat 0·07, bran 0·14, white flour 0·0052 $\mu g.$/g. (Lampen *et al.* 1942). From this the biotin content of wheat would be optimal for *Tribolium*, but of white flour near the threshold. A casein-glucose diet, with 5% yeast, as used in many of our experiments, would contain 0·04 $\mu g.$/g. from yeast alone, which is likely to be sufficient for *Tribolium*.

Summary.

Insects when bred on an artificial diet consisting of casein, glucose, cholesterol, salts and aqueous yeast extract require in addition, for optimal growth, the water-insoluble residue of yeast. The "insoluble factor" is not fat-soluble, and becomes soluble on autolysis or by autoclaving. Experiments are described which led to its recognition as biotin.

We gratefully acknowledge a Government grant, made by the Royal Society, which, partly, made this investigation possible.

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Book Notices.

Laboratory Procedures in Studies of the Chemical Control of Insects. Ed. F. L. Campbell and F. R. Moulton. *Publ. Amer. Ass. Adv. Sci.* **20** : 1–206, text illust., 1943. Washington.

This volume is the result of a meeting of the American Association of Economic Entomologists and the American Association for the Advancement of Science held in 1941, and most of the papers were read at that meeting, although many have since been revised and others re-written.

The volume presents " a systematic, comprehensive, authoritative, and thoroughly documented discussion " arranged in six sections : (1) Introduction, (2) Rearing test insects, (3) Methods of testing insecticides against insects in the laboratory, (4) Statistical methods, (5) Bibliography, and (6) Indices in which both the scientific and common names of the insects mentioned are given.

Keys to the British species of Ephemeroptera, with keys to the genera of the nymphs. By D. E. Kimmins. *Sci. Publ. Freshwater biol. Ass. British Empire* **7** : 1–64, text illust., 1942. Price 2*s*. 6*d*.

This pamphlet opens with an introduction on the position of the Ephemeroptera in the classification of insects, followed by some general remarks on their occurrence in Britain. A revised check list of the British species is given and also some notes on collection and preservation, and a list of the names by which the several " Mayflies " are known to fly-fishermen. The systematic part opens with a key to the families, followed by a list of all the genera, and an indication of the species known to occur in this country. Excellent drawings by the author illustrate the book, which is attractively produced, and it is completed by a bibliography and a short index.

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A uniform method is adopted for the citation of bibliographical references in the Society's publications as follows:

Smith, A., 1936, New species of Coccidae, *Proc. R. ent. Soc. Lond.* (B) **6** : 301–306, pl. 1.

——. 1936, New species of Coccidae, *Trans. R. ent. Soc. Lond.* **84** : 901–936.

Titles of periodicals cited are to be abbreviated in the manner indicated in the *World List of Scientific Periodicals, 2nd edition*, 1934.

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41, Queen's Gate, S.W.7

1944

WEDNESDAY,	April	5
,,	May	3
,,	June	7
,,	October	4
,,	November	1
,,	December	6

1945.

WEDNESDAY,	January	17 (ANNUAL MEETING)
,,	February	7

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